

## WHAT IS CLAIMED IS:

1. A method for the determination of lipid individual molecular species composition of matter and amount by algorithm analysis in a biological sample comprising subjecting the biological sample to lipid extraction to obtain a lipid extract and subjecting the lipid extract to two dimensional (or multidimensional) (or multi-dimensional) electrospray ionization tandem mass spectrometry (ESI/MS/MS) by iterative processing producing the determination of structure and amount.
2. A method in accordance with Claim 1 wherein the lipid extraction is a chloroform lipid extraction.
3. A method in accordance with Claim 2 wherein said extraction is of a blood, serum, tissue biopsy, feces and urine sample.
4. A method in accordance with Claim 3 wherein said TG biological sample is one of a mammalian tissue and a plant tissue.
5. A method in accordance with Claim 4 wherein the mammalian tissue is human tissue and the lipid is a triacylglyceride.
6. A method in accordance with Claim 5 wherein the determination comprises a finger print profile of a patient's triglyceride molecular species and provides a quantitative analysis of individual species.
7. A method in accordance with Claim 6 wherein said finger profile comprises the individual molecular species of a triglyceride composition of matter.
8. A method for the determination of lipid individual molecular species composition of matter directly from a lipid extract of a biological sample comprising subjecting said lipid extract to electrospray ionization tandem mass spectrometry.

9. A method in accordance with Claim 1 wherein said lipid extraction is chloroform extraction and the lipid is a triacylglyceride.

10. A method in accordance with Claim 9 wherein said TG biological sample is one of a mammalian or a plant tissue.

11. A method in accordance with Claim 10 wherein said mammalian tissue is human tissue.

12. A method in accordance with Claim 6 wherein the biological sample is an aqueous human fluid sample subjected to centrifugation and/or conventional column chromatography suitable for separation of lipoproteins to resolve lipids into different lipoprotein fractions.

13. A method in accordance with Claim 6 wherein the aqueous human fluid sample is selected from the group consisting of whole blood, blood serum, blood plasma, liver and urine.

14. A method in accordance with Claim 13 wherein the lipid extract is obtained by extraction of said biological sample with chloroform.

15. A method in accordance with Claim 14 wherein the triglyceride molecular species of the biological sample are determined by comparison with the triglyceride molecular species of a standard control sample.

16. A method in accordance with Claim 15 wherein the triacylglyceride molecular species of the biological sample are determined by comparisons of their ion peak intensities with the ion peak intensities of a standard control sample and iteratively deconvoluted and optionally normalized to yield quantitative information on mass of molecular species.

17. A method in accordance with Claim 16 wherein said determination includes deconvolution of the intensity two dimensional (or multidimensional) or multidimensional intercept contours of the triglycerides at their neutral loss products.

18. A diagnostic kit for the determination of lipid molecular species in a biological sample comprising components suitable for carrying out any of the method of Claim 1 and quantitative determinations described therein.

19. A method for assessing a risk to an individual based on TG molecular species as an independent factor in the development of at least one condition in that individual for a medical condition selected from coronary artery disease, stroke, atherosclerosis and obesity which comprises analyzing a biological sample taken of an individual for TG molecular species determination, administering a therapeutic amount of a drug to the individual (treated), analyzing a corresponding biological sample of the treated individual, comparing the TG molecular species determination after drug administration with the TG molecular species determination prior to the drug administration and determining the benefit of decreased risk due to the drug now afforded to that individual.

20. A method in accordance with Claim 19 wherein the comparison of the TG molecular species determination of the biological samples is indicative of development of the condition for that individual.

21. A method for identifying an agent which selectively targets specific to lipid or triacylglyceride molecular species (e.g., saturated triacylglycerides) which comprises analyzing a biological sample of at least one treated individual for TG molecular species determination, administering a drug to the individual, analyzing a biological sample from said treated individual, comparing the TG molecular species determination after said administration with the TG molecular species determination prior to drug administration and determining an effect on the treated individual of the drug administration.

22. A method in accordance with Claim 21 wherein said comparison of the TG molecular species determination of the biological samples is indicative of development of the condition for that individual.

23. A method of identifying a candidate lipid modulating drug having lipid modulating drug efficacy which comprises analyzing a biological sample

of at least one individual subject for TG molecular species determination, administering a therapeutic amount of a candidate lipid modulating drug to the individual subject, analyzing a biological sample of said administered individual, comparing the TG molecular species determination after said administration with the TG molecular species determination prior to the drug administration and determining an effect if any on the individual of the drug administration.

24. A method in accordance with Claim 23 wherein said comparison of TG analysis is indicative of a lipid modulating capacity of an administered drug.

25. A method in accordance with Claim 24 wherein said modulating comprises lowering.

26. A method for diagnosing and determining the response of a patient to tailored drug therapy which comprises analyzing a biological sample of a patient to be treated or TG molecular species determination, administering amount of a drug to the patient, analyzing a biological sample taken from the treated patient, comparing the TG molecular species determination after the administration with the TG molecular species determination prior to the drug administration and determining an effect on the treated patient of the drug administration.

27. A method in accordance with Claim 26 wherein said comparison of TG analysis is indicative of a successful tailored drug therapy.

28. A method of screening candidate chemicals for lipid modulating efficacy in a subject which comprises analyzing a biological sample of a subject for TG molecular species determination, administering a therapeutic amount of a drug to that biological subject, analyzing a biological sample of said subject, comparing the TG molecular species determination after said administration with the TG molecular species determination prior to the drug administration and determining an effect if any on the subject of the drug administration.

29. A method of screening in accordance with Claim 28 wherein said comparison of TG analysis is indicative of a candidate chemical having a lipid lowering potential on a human subject.

30. A method of treating a subject comprising analyzing a biological sample taken of that subject for lipid i.e. TG molecular species determination by the method by multidimensional ESI/MS and quantitative changes.

31. A medical treatment in accordance with Claim 30 wherein the subject is a living human.

32. A medical treatment in accordance with Claim 31 wherein said treatment is medicinal and therapeutic.

33. A medical treatment comprising analyzing a biological sample taken of a subject for TG molecular analysis determination by multidimensional ESIMS and prescribing a therapy based on the determination.

34. A medical treatment in accordance with Claim 33 wherein the subject is a human.

35. A method in accordance with Claim 34 wherein said medical treatment is therapeutic.

36. A method of customizing drug therapy lipid i.e. a subject which comprises analyzing a biological sample taken of the subject for TG molecular species determination by multidimensional ESI/MS and customizing the subject's drug therapy based on the results of the TG molecular species determination and quantitative changes.

37. A method of customizing drug therapy in accordance with Claim 34 wherein the subject is human.

38. A method of retarding, preventing, ameliorating or diagnosing disease in a subject based on lipid i.e. TG molecular species determination of a biologic sample of the subject, which comprises analyzing a biological sample taken

of a subject for TG molecular analysis by multidimensional ESIMS determination associated with the disease and prescribing a therapy for the subject based on the TG molecular species determination.

39. A method in accordance with Claim 38 wherein the subject is human.

40. A method of managing a library of chemicals which comprises administering a chemical selected from the library to a subject and analyzing a biological sample taken of that subject for lipid. i.e. TG molecular species determination by multidimensional ESI/MS, quantitating the mass of individual entities and assigning a priority to said chemical for further development based on that determination.

41. A method in accordance with Claim 40 wherein the subject is human.

42. A method of determining a subject's response to administration of a drug which comprises administering a drug to the subject, and analyzing a biological sample taken of a subject for lipid i.e. TG molecular analysis by multidimensional ESI/MS following said administration, molecular species and quantitation.

43. A method in accordance with Claim 42 wherein the subject is human.

44. A method of providing a medical assessment to a subject which comprises analyzing a biological sample taken of a subject for lipid i.e. TG molecular analysis by multidimensional ESI/MS and providing an assessment to the subject based on that determination.

45. A method of providing a medical assessment in accordance with Claim 44 wherein the subject is human.

46. A method of enhancing medical care provided to a subject which comprises analyzing a biological sample taken of a subject for TG molecular analysis by multidimensional ESI/MS and providing a modulated therapy to the subject.

47. A method of enhancing subject care in accordance with Claim 46 wherein the subject is human.

48. A method in accordance with Claim 47 wherein the TG molecular analysis is a TG molecular analysis of species and simultaneous quantitation.

49. In an aspect, a method is provided to identify and quantify multiple lipid species concurrently directly from their lipid extracts of biologic samples through intrasource separation and multidimensional analysis of mass spectra from precursor ion and neutral loss scans of naturally occurring lipid fragments.

50. In an aspect, multidimensional analysis of samples which are subject to derivatizations to those skilled in the art such as derivatization of primary amines (aldehydes and other agents), double bonds (dimethyldisulfide, diborane or other common reagents), sugars, phosphates, primary hydroxyl (trimethylsilyl chloride) and other common derivatizing agents.

51. In an aspect, a ratiometric comparison of lipids between two states (e.g., control and disease) is carried out by derivatization with light and heavy isotopes to determine the relative amounts of each molecular species after multidimensional mass spectrometric analysis by these methods.

52. In an aspect, a method for identification of biomarkers of disease, prognostic indicators of disease outcome or markers of treatment efficacy in disease states which can be identified through multidimensional mass spectrometry by a systems biology bioinformatics approach which is provided by correlating the mass of lipid products and metabolites with disease onset, severity or progression.

53. In an aspect, this methodology of this discovery encompasses a method for an automated platform for multidimensional lipid analysis capable of analyzing thousands of different lipids through multidimensional mass spectrometry through commonly employed principles of automation (e.g., automated sample injection) and data analysis (e.g., deisotope deconvolution) as routinely employed by those skilled in the field.

54. In an aspect, this discovery of a multidimensional mass spectrometry provides a means for obtaining abundant novel chemical information about spatial relationships in lipid molecules (e.g., regiospecificity, chemical linkages and relative abundance of isobaric and other species) not accessible by the one dimensional approach.

55. A method in accordance with Claim 1 wherein said lipid comprises at least one of phospholipids (e.g., choline glycerophospholipides (e.g., plasmenycholine, phosphatidylcholine, plasmanylcholine), sphingomeyelin, ethanolamine glycerophospholipids, mono and dimethyl ethanolamine, glycerophospholipds, serine glycerophospholipids, inositol glycerophospholipids, cardiolipin, phosphatidic acid, phosphatidylglycerol, phasphatidylethanol and oxidized derivatives thereof), fatty acids, fatty amides, eicosanoids, sphingolipids, glycolipids, steroids, ceramides, acylCoA, acylcarnitine, acylprotiens, acylpeptides, diglycerides, monoglycerides, anadamide and 2-arachidonyl glycerol or oxidized nitrated or sulfated species therefrom or other derivatives know to those in the field.

56. A method in accordance with Claims 19, 20 and 23 wherein said lipid comprises at least one of phospholipids (e.g., choline glycerophospholipides (e.g., plasmenycholine, phosphatidylcholine, plasmanylcholine), sphingomeyelin, ethanolamine glycerophospholipids, mono and dimethyl ethanolamine, glycerophospholipds, serine glycerophospholipids, inositol glycerophospholipids, cardiolipin, phosphatidic acid, phosphatidylglycerol, phasphatidylethanol and oxidized derivatives thereof), fatty acids, fatty amides, eicosanoids, sphingolipids, glycolipids, steroids, ceramides, acylCoA, acylcarnitine, acylprotiens, acylpeptides, diglycerides, monoglycerides, anadamide and 2-arachidonyl glycerol.